

**Amendments to the Claims:**

Please add the following new Claims 31-33 to this application. This listing of claims will replace all prior versions and listings of the claims in the application:

1. (original) A reaction mixture for performing protein synthesis reaction, the mixture comprising a prokaryotic S-30 extract combined with a supplemental mix containing buffer, salts, nucleotide triphosphates, an energy generating system, and amino acids, the reaction mixture being substantially depleted in RNase E.
2. (original) A reaction mixture as claimed in Claim 1 wherein the extract is from *E. coli*.
3. (original) A reaction mixture as claimed in Claim 1 wherein the mixture further comprises an amount of amino acids.
4. (original) A reaction mixture for performing protein synthesis reactions, the mixture comprising a prokaryotic S-30 extract combined with a supplemental mix containing buffer, salts, nucleotide triphosphates, an energy generating system, the reaction mixture having the degradosomes substantially removed therefrom.
5. (original) A reaction mixture as claimed in Claim 4 wherein the extract is from *E. coli*.
6. (original) A reaction mixture as claimed in Claim 4 wherein the mixture further comprises an amount of amino acids.
7. (original) A reaction mixture for performing protein synthesis reactions, the mixture comprising a prokaryotic S-30 extract combined with a supplemental mix containing buffer, salts, nucleotide triphosphates, and an energy source, wherein the reaction mixture had been fractionated by freezing, thawing and centrifugation.
8. (original) A reaction mixture as claimed in Claim 7 wherein the extract is from *E. coli*.

9. (original) A reaction mixture as claimed in Claim 7 wherein the mixture further comprises an amount of amino acids.

10. (withdrawn) A protein synthesis reaction mixture comprising a combination of an S-30 extract and supplemental mix that has been fractionated by freezing, thawing and centrifugation.

11. (withdrawn) An article of manufacture comprising  
a fractionated *E. coli* S-30 reaction mixture which is composed of the combined constituents of an S-30 extract and a supplemental mix combined and fractionated, the fractionation removing RNase E from the mixture; and  
a container suitable for storage and shipment containing the fractionated S-30 reaction mixture.

12. (withdrawn) An article of manufacture as claimed in claim 11 wherein the reaction mixture is frozen.

13. (withdrawn) An article of manufacture as claimed in claim 11 wherein the reaction mixture is dried.

14. (withdrawn) An article of manufacture as claimed in claim 11 wherein the S-30 reaction mix was made by the process of combining an S-30 extract and a supplemental mix to make a cloudy solution followed by centrifugation of the solution, saving the supernatant.

15. (withdrawn) An article of manufacture comprising  
a fractionated *E. coli* reaction mixture which is made by combining an S-30 extract and a supplemental mix and then fractionating the combination, the fractionation removing most of the DNA from the mixture; and  
a container suitable for storage and shipment containing the fractionated reaction mixture.

16. (withdrawn) An article of manufacture as claimed in claim 15 wherein the reaction mixture is frozen.

17. (withdrawn) An article of manufacture as claimed in claim 15 wherein the reaction mixture is dried.

18. (withdrawn) An article of manufacture as claimed in claim 15 wherein the S-30 reaction mix was made by the process of combining an S-30 extract and a supplemental mix to make a cloudy solution followed by centrifugation of the solution, saving the supernatant.

19. (withdrawn) An article of manufacture comprising  
a fractionated *E. coli* reaction mixture which is made by combining an S-30 extract and a supplemental mix and then fractionating the combination, the fractionation having the RNA degradosomes from the *E. coli* substantially removed;  
and

a container suitable for storage and shipment containing the fractionated reaction mixture.

20. (withdrawn) An article of manufacture as claimed in claim 19 wherein the reaction mixture is frozen.

21. (withdrawn) An article of manufacture as claimed in claim 19 wherein the reaction mixture is dried.

22. (withdrawn) An article of manufacture as claimed in claim 19 wherein the S-30 reaction mix was made by the process of combining an S-30 extract and a supplemental mix to make a cloudy solution followed by centrifugation of the solution, saving the supernatant.

23. (withdrawn) A method of making a reaction mixture for conducting a protein synthesis reaction in a prokaryotic cell free extract, the method comprising the steps of

- (a) making an *E. coli* S-30 extract by lysing *E. coli* cells and centrifuging the lysate;
- (b) separately, before or after step (a), making a supplemental mix including buffer salts, nucleotide triphosphates, an energy generating system, and precipitating agent that preferentially precipitates high molecular weight molecules;
- (c) combining the solutions of step (a) and (b); and
- (d) centrifuging the combined solutions and separating the supernatant to make the reaction mixture.

24. (withdrawn) A method as claimed in claim 23 wherein the precipitating agent is polyethylene glycol.

25. (withdrawn) A method as claimed in claim 23 wherein after step (c) the combined solutions are frozen and thawed prior to the centrifuging of step (d).

26. (withdrawn) A method as claimed in claim 23 further comprising the steps of placing the supernatant into containers for commercial sale.

27. (withdrawn) A method of performing an *in vitro* protein synthesis reaction, the method comprising the steps of

(a) making an *E. coli* S-30 extract by lysing *E. coli* cells and centrifuging the lysate;

(b) separately, before or after step (a), making a supplemental mix including buffer, salts, nucleotide triphosphates, an energy generating system, and a precipitating agent that acts to preferentially precipitate high molecular weight components;

(c) combining the solutions of step (a) and (b);

(d) centrifuging the combined solutions and separating the supernatant to make the reaction mixture;

(e) adding a DNA template to the reaction mixture, the DNA template encoding the expression of a protein and including a promoter recognized by an RNA polymerase in the reaction; and

(f) incubating the mixture under conditions such that protein is produced.

28. (withdrawn) A method as claimed in claim 27 wherein the precipitating agent is polyethylene glycol.

29. (withdrawn) A method as claimed in claim 27 wherein after step (c) the combined solutions are frozen and thawed prior to the centrifuging of step (d).

30. (withdrawn) A method as claimed in claim 27 further comprising the steps of placing the supernatant into containers for commercial sale.

31. (new) A reaction mixture for performing protein synthesis reactions, the reaction mixture comprising an *E. coli* S-30 extract combined with a supplemental mix containing buffer, salts, nucleotide triphosphates, and an energy source,

the S-30 extract prepared by the process of

- (a) growing *E. coli* cells in culture;
- (b) centrifuging the cells to make a cell pellet;
- (c) resuspending the cell pellet;
- (d) pressing the resuspended cell pellet in a French press to lyse the

cells; and

- (e) centrifuging the lysed cells, the supernatant being the S-30 extract;

wherein the reaction mixture, after preparation, is further processed by the step of fractionating the reaction mixture by freezing, thawing and centrifugation.

32. (new) A reaction mixture for performing protein synthesis reactions, the mixture comprising *E. coli* transcriptional and translational components combined with a supplemental mix containing buffer, salts, nucleotide triphosphates, an energy generating system, the *E. coli* transcriptional and translational components being made up of the material which is in the supernatant of lysed and centrifuged *E. coli* cells, the reaction mixture having the degradosomes substantially removed therefrom.

33. (new) A reaction mixture for performing protein synthesis reactions, the mixture comprising *E. coli* transcriptional and translational components combined with a supplemental mix containing buffer, salts, nucleotide triphosphates, an energy generating system, the *E. coli* transcriptional and translational components by the process of

- (a) growing *E. coli* cells in culture;
- (b) centrifuging the cells to make a cell pellet;
- (c) resuspending the cell pellet;
- (d) pressing the resuspended cell pellet in a French press to lyse the

cells; and

- (e) centrifuging the lysed cells, the supernatant being the S-30 extract;

the reaction mixture having the degradosomes substantially removed therefrom by the process of freezing thawing and centrifuging the combined reaction mixture.